Monkeypox and whitepox viruses in West and Central Africa*

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Prospects for the eradication of smallpox are now highly encouraging. With the cessation of man-to-man transmission, the question of possible animal reservoirs of smallpox becomes increasingly important. During the period 1970–1975, 20 cases of a smallpox-like disease were detected in smallpox-free areas of tropical rain forest in West and Central Africa. Epidemiological and virological investigations revealed that the disease was caused by an animal poxvirus termed monkeypox virus, a member of the orthopox virus group. The disease spread with difficulty even among susceptible close contacts and does not appear to be sufficiently transmissible to permit continuing infection to become established in man. During the investigations, four orthopox viruses termed whitepox viruses were isolated from rodents and monkeys. The isolates were not distinguishable from variola virus with currently available laboratory techniques, but there is no evidence so far that viruses of this group have infected man. Although there is now substantial and accumulating evidence that there is no animal reservoir for smallpox, continued surveillance and studies in West and Central Africa are warranted.

Smallpox eradication has been defined by the WHO Expert Committee on Smallpox Eradication (1) as "...the elimination of clinical illness caused by variola virus." The Committee added: "... since there is no human carrier state of epidemiological importance and no recognized animal reservoir of the disease, the absence of clinically apparent cases in man may be assumed to signify the absence of naturally occurring smallpox." Evidence of the absence of an animal reservoir (2) has been strengthened over the last ten years by the fact that greatly intensified epidemiological surveillance has not documented a single outbreak of smallpox in a smallpox-free area except when introduced by man from a known smallpox-infected area.

During the past ten years, however, monkeypox virus, previously thought to be restricted to monkey populations, has been recovered from 20 cases in man of an exanthematous disease clinically indistinguishable from smallpox. During epidemiological investigation of these cases, all of which occurred in

West and Central Africa, four isolates resembling variola virus were obtained from specimens collected from monkeys and rodents captured in the area where the cases had occurred. These four strains, along with two strains isolated in Utrecht (3) from kidneys of healthy monkeys, have been termed "whitepox viruses".

MONKEYPOX

Monkeypox outbreaks in captive monkey colonies

Since 1958, when the first monkeypox outbreak in a captive monkey colony was reported (4), nine additional outbreaks in laboratories have been recorded (four in Europe and five in the USA) (5). Six of the ten outbreaks were confirmed by the isolation of monkeypox virus. At least four occurred among monkeys shipped from Malaysia. The significance of this is uncertain, however; of 481 sera obtained from monkeys in Malaysia (5), none possessed poxvirus antibody. It is therefore quite possible that susceptible Malaysian monkeys acquired infection from other simians or mammals during shipment to the laboratories.

The last known outbreak of monkeypox occurred in a laboratory in Paris in 1968, the disease having

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occurred in one of two chimpanzees shipped from Sierra Leone where a case of monkeypox in man was detected in 1970 (6). Despite the increased alertness of laboratories to the possible occurrence of monkeypox, no outbreak has been reported since then. Furthermore, no naturally occurring monkeypox outbreaks have been documented in wild monkey populations.

Monkeypox in man

In 1970, the first known case of monkeypox in man was discovered in a tropical rain forest area in Equateur Province, Zaire (7). In the same year, five cases were found in Liberia and Sierra Leone (8). From 1970 to December 1975, a total of 20 cases in man were recorded in West and Central Africa (Table 1).

Of the 20 patients with monkeypox virus infection, 13 were children under the age of 5 years, two were between 6 and 15 years of age, and the remaining 5 were adults. None of the cases, except case 6 (24 years old) and case 12 (30 years old) had been vaccinated against smallpox before the presumed date of exposure. The clinical disease in most patients resembled smallpox and 4 of the 20 patients died. Among the 8 cases that occurred in Sierra Leone, Liberia, Ivory Coast, and Nigeria, 3 patients were left with distinct facial pockmarks that persisted after 5 years.

Laboratory diagnosis. In 13 cases, the diagnosis of monkeypox was confirmed by the isolation of monkeypox virus, which is distinguishable from variola virus but belongs to the orthopox group (4, 9, 10). In the remaining seven cases, the diagnosis was made by the detection of poxvirus by electron microscopic examination and/or by the presence of poxvirus antibody in the sera. Notably, in cases 7 and 8 (Nigeria) and 9 (Ivory Coast), sera taken five years after the onset of the disease showed specific monkeypox antibody (11).

Transmission. The 20 cases occurred in 15 different villages. Eleven cases occurred as single-case outbreaks. Cases 2, 3, and 4 occurred in village B, Liberia, within 2 days of each other, suggesting that the patients had been exposed to infection at almost the same time. Cases 7 and 8 occurred in the same family in village E, Nigeria. Case 8, the mother of case 7, developed a mild rash 9 days after her child's rash began, suggesting that man-to-man transmission may have occurred. Cases 11 and 12 occurred in village H, Zaire on the same day, implying common

source exposure. Cases 15 and 16 occurred in village K, Zaire with an interval of 12 days. Case 15 is the sister of case 16, which again suggests that manto-man transmission may have occurred.

On only two occasions, therefore, is it believed that monkeypox may have been transmitted from man to man. Notably, in the Nigeria outbreak, there were 12 unvaccinated family members in contact with the patient but only one (case 8) became infected. Review of all known outbreaks to date reveals that only 2 of 30 susceptible household contacts (6.6%) developed the disease, a transmission rate far lower than the 35% observed in smallpox outbreaks (12, 13).

Source of infection. In each locality (coded A to O, Table 1) and in the surrounding area, extensive epidemiological investigations were carried out to seek the source of monkeypox infection in man. These investigations have failed to detect any smallpox or monkeypox cases before or after the occurrence of the index cases, except for the secondary household cases 8 and 16 described above. Particular attention was paid to the history of contact with monkeys, which are abundant in the areas surrounding all the localities. Cases 7, 10, 11, 14, 17, and 20 had had close contact with monkeys before illness developed. Each had either eaten monkey meat, prepared monkeys for cooking, or played with live or dead monkeys. However, it has not yet been possible to obtain suspected source material for laboratory investigation.

All the localities where cases occurred are situated in tropical rain forest, except for locality M, which is a town with a population of 40 000. Case 18 occurred in the centre of this town, and there was no history of the patient having travelled outside the town before illness developed. On three occasions 1, 6, and 12 months after this outbreak, special searches were conducted in primary schools to detect children with facial pockmarks. A total of 94 children with facial scarring caused by disease during the previous three years were found, including case number 18. All showed vaccination scars, except case number 18 and one other. Clinical and epidemiological investigations indicated that the facial scarring had been produced by chickenpox (E. Zanotto, personal communication, 1975).

In 1975, a special survey was conducted in Sierra Leone, Liberia, the Ivory Coast, and Nigeria, almost five years after smallpox cases last occurred in these countries, in order to detect any additional monkey-

Table 1. Cases of monkeypox virus infection in man

Case No.	Loca- lity	Province	Country		Sex	Vac- cin- ation scar	Date of onset of rash	Sev- erity ^a	Death	Laboratory results			
				Age (years)						Poxvirus (elec- tron micro- scopy)	Monkey- pox isolate	Pox antibody	Specific monkey- pox antibody
1	Α	Equateur	Zaire	9 months	М	_	24.8.70.	2	_		+		
2	В	Grand Gedah	Liberia	4	F	_	12.9.70.	2	_	+	+	+	
3	В	Grand Gedah	Liberia	4	М	_	13.9.70.	1				+	
4	В	Grand Gedah	Liberia	6	F	_	13.9.70.	1				+	
5	С	Grand Gedah	Liberia	9	М		2.10.70.	2		+	+	+	
6	D	Aguebu	Sierra Leone	24	М	+	1.12.70.	2	_	+	+	+	
7	Ε	Aba	Nigeria	4	F	_	9.4.71.	3	_	+	+		+
8	E	Aba	Nigeria	24	F	_	18.4.71.	1	_				+
9	F	Aben Gourrou	Ivory Coast	5	М	_	18.10.71.	2	_			+	+
10	G	Kasai Oriental	Zaire	1	М		2.3.72.	2	-	+	+		
11	Н	Equateur	Zaire	3	М		27.7.72.	3	+	+	+		
12	Н	Equateur	Zaire	30	F	+	27.7.72	1	_			+	
13	1	Equateur	Zaire	7 months	F	-	16.9.72.	2	+	+			
14	J	Bandundu	Zaire	2	М		30.10.72.	2	+	+	+		
15	Κ	Equateur	Zaire	3	F	_	10.1.73.	2	_	+	+	+	
16	Κ	Equateur	Zaire	5	F	-	22.1.73.	2	_	+		+	
17	L	Equateur	Zaire	7 months	М		6.5.73.	3	+	+	+		
18	М	Equateur	Zaire	4	F	_	6.8.74.	2	_	+	+		
19	N	Bandundu	Zaire	40	F	_	4.1.75.	3	_		+	+	
20	0	Kasai Oriental	Zaire	23	F	_	9.3.75.	1	_	+	+	+	

a 1 = mild; 2 = intermediate; 3 = severe.

pox cases. The survey teams searched the villages where the cases had occurred and the surrounding 10–30 villages (I. D. Ladnyi & R. Netter, personal communications, 1976). These studies failed to detect any additional cases despite the fact that only 40–70% of the village populations had been vaccinated.

SEARCH FOR AN ANIMAL RESERVOIR OF MONKEYPOX

Epidemiological investigations of monkeypox cases in man have failed to trace the source. As noted above, a serological survey of 481 monkeys in Malaysia revealed none with poxvirus antibody. Additionally, of 1614 monkey sera obtained from

various African and Asian sources (survey 1), none had significant poxvirus antibody titres (5) (Table 2).

However, serological surveys conducted in areas of West and Central Africa, where monkeypox infection of man had occurred, revealed the presence of haemagglutinin inhibiting and neutralizing antibodies in a number of animals (surveys 3–11). Since it is not possible at present to determine which orthopox virus infection might have induced antibody production, and since there are many different naturally occurring poxviruses in mammals, the significance of these observations is unknown. Notably, in two sera from survey 7, Gispen (11) has reported the presence of specific monkeypox antibody as measured by immunofluorescence. Both sera were collected from adult female Cercopithecus

Table 2.	Investigations	of wild	animals in	relation to	o monkevpox	outbreaks in man
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Survey No.	Testing laboratory	Year of specimen collection	Place of specimen collection	No. of specimens	Animals	Type of specimens	POXVIPUS	Circumstances for collection of specimens random	
1	Monkeypox Study Group ^a	collected in 1963-69 but tested in 1970-71	Japan Philippines, In- donesia, Malay- sia, Thailand India Chad, Upper Volta, Mali, Kenya, Senegal unknown total	378 165 703 304 1614	monkeys	serum	-		
2	Monkeypox Study Group ^a	1970	Malaysia	481	monkeys	serum		random	
3	Moscow	1970	Zaire	9	monkeys	serum & kidney	white poxvirus (chimp 9)	case 1	
4	Atlanta	1971	Liberia	371	monkeys, rodents, and other miscellaneous species	serum and various tissues	no isolate	cases 2–4	
5	Atlanta	1971	Nigeria	68	monkeys and rodents	serum and various tissues	no isolate	cases 7–8	
6	Atlanta	1971–72	Ivory Coast ^b	102	monkeys, rodents, and other miscellaneous species	serum	_	case 9	
7	Atlanta, Utrecht	1973–74	Ivory Coast ^b	207	monkeys	serum	_	case 9	
8	Moscow	1973	Zaire	12	monkeys	serum & kidney	white poxvirus (MK-7-73)	cases 15-16	
9	Moscow	1973	Zaire	92	monkeys & rodents	serum & kidney	no isolate	case 17	
10	Moscow	1974	Zaire	378	monkeys & rodents	serum & kidney	white poxviruse (RZ-10-74, RZ-38-75)	s case 18	
11	Moscow	1975	Zaire	67	monkeys & rodents	serum & kidney	no isolate	case 19	

^a Monkeypox Study Group: R. Gispen, Rijks Instituut voor de Volksgezondheid, Utrecht, Netherlands; S. S. Kalter, Southwest Foundation for Research and Education, San Antonio, Texas, USA; Lim Teong Wah, Institute for Medical Research, Kuala Lumpur, Malaysia; S. S. Marennikova, Research Institute of Virus Preparations, Moscow, USSR; R. Netter, Laboratoire national de la Santé publique, Paris, France; and I. Tagaya, National Institute of Health, Tokyo, Japan.

aethiops monkeys in the northwestern part of the Ivory Coast in 1973 (J. Breman, personal communication, 1976). Further studies are in progress.

Efforts to isolate monkeypox virus from specimens collected in these surveys were unsuccessful. However, in surveys 3, 8, and 10 conducted in Zaire, the Research Institute of Virus Preparations, Moscow, USSR recovered four variola-like poxvirus isolates. The isolates cannot be distinguished from variola virus by currently available laboratory tests. All produce small, whitish pocks on the chorioallantois of chick embryos, similar to those produced by variola virus. For purposes of reference, they have

been termed "whitepox viruses". A whitepox virus strain, inoculated experimentally, caused generalized rash in *C. aethiops* monkeys (J. Nakano, personal communication, 1976). Despite the large number of specimens collected in surveys 4 and 5 in Liberia and Nigeria, no isolate was obtained.

WHITEPOX VIRUS

All four whitepox viruses have been isolated from monkey or rodent specimens collected in Equateur Province, Zaire, where eight cases of monkeypox in man have been detected. The district consists of

^b Serum samples supplied by Dr J. Breman, Organization for Coordination and Cooperation in the Control of Major Endemic Diseases (OCCGE), Bobo-Dioulasso, Upper Volta.

dense tropical rain forest. There is a rainy season from February to November but even during the "dry" months of December and January it usually rains 2 or 3 times a week. The fauna of the region is very varied; the species of monkey seen most frequently are chimpanzees, *Colobus*, *Cercopithecus*, mangabey and, in the north, baboons. Among the rodents, there are numerous species of squirrels and rats, and porcupines.

The circumstances in which the viruses were isolated are summarized in Table 3. The strains are as follows:

Chimp 9

This strain was isolated from the kidney of a chimpanzee captured and killed near locality A, Basankusu area, where the first case of monkeypox in man (No. 1) was detected (10). The investigation during which this chimpanzee specimen was collected was made four months after the case occurred. Serum was also collected from the same chimpanzee and showed a significant poxvirus antibody titre, indicating that the animal had experienced a poxvirus infection. However, no poxvirus was isolated from specimens collected from eight other monkeys, although some of them showed positive poxvirus antibody titres.

MK-7-73

This strain was isolated from the kidney of an unidentified monkey captured during investigation in Ubangi area, Equateur Province, where cases 15 and 16 had occurred. Again poxvirus antibody was detected in the serum of the same animal. Specimens from eleven other monkeys were tested with negative results (S. S. Marennikova, personal communication, 1973).

RZ-10-74 and RZ-38-75

These strains were isolated from the kidneys of two rodent species (Mastomys natalensis and Heliosciurus rufobrachium) captured in Bumba zone, Equateur, during investigation of case 18 (S. S. Marennikova, personal communication, 1976). Notably, in Bumba zone between 1972 and 1975 four human monkeypox cases (11, 12, 17, and 18) occurred within an area of 100 km radius. RZ-10-74 was isolated from a specimen collected one month after case 18 had occurred. This virus was repeatedly isolated from the original specimen, but no sera were available for serological confirmation. RZ-38-75 was isolated from a specimen collected eight months after the occurrence of case 18. The serum of the rodent from which RZ-38-75 was isolated showed poxvirus antibody. On these two occasions, virus isolation tests on a total of 48 monkey specimens and 328 rodent specimens were negative. Studies of antibody titres in these sera are in progress.

LABORATORY WHITEPOX VIRUS

In September 1964, during routine processing of Malaysian cynomolgus monkey kidney tissue cultures at the Rijks Instituut voor de Volksgezondheid,

Table 3.	Wild	whitepox	virus	isolates	trom	Equateur	Province, Z	Laire
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						Confirmation of isolation			
Survey No.	Strain	Isolated from:	Place of specimen collection	Date of onset of monkeypox in man	Date of specimen collection	pox antibody titre in serum from original animal		repeat	
						HAI	neutral- izing	isolation	
3	Chimp 9	chimpanzee	near locality A, Basankusu	August 1970 (case 1)	January 1971	1280	40	ND	
8	MK-7-73	" sala "a monkey	near locality K, Ubangi	January 1973 (cases 15-16)	February 1973	256	80	ND	
10	RZ-10-74	Mastomys natalensis	near locality M, Bumba Town	August 1974 (case 18)	September 1974	ND ^b	ND	+	
10	RZ-38-75	Heliosciurus rufobrachium	100 km north of locality M, Bumba Town	August 1974 (case 18)	March 1975		80	ND	
						_	20		

a Local name; it was not possible to identify the species.

b ND = not done.

Utrecht, Netherlands, poxviruses were isolated on two occasions (specimens 64-7275 and 64-7255) (14). These two isolates were found to resemble variola virus and were designated "whitepox viruses". No similar strains have been isolated before or since.

Of uncertain relevance is the fact that on 21 December, three months after the isolation of the two whitepox virus strains, anteaters that had arrived at Rotterdam Zoo on 9 December developed a vesicular disease. Monkeypox virus was isolated from these animals (15). Investigation revealed that the animals, prior to the development of disease, had had close contact with cynomolgus monkeys that had probably been shipped from Malaysia. These anteaters were the source of a subsequent monkeypox outbreak in the Rotterdam Zoo, in which 21 monkeys and apes of various species suffered from the disease and 11 died (16).

On 9 December 1964 and 4 May 1965, the laboratory in Utrecht again isolated poxviruses from tissue culture of cynomolgus monkey kidney. This time, however, the isolates resembled monkeypox virus (specimens 64-9411 and 65-3993).

The evidence suggests a common focus for the monkeypox virus strains and the whitepox viruses detected in the Utrecht laboratory, as well as for the monkeypox outbreak at Rotterdam Zoo.

DISCUSSION

The last smallpox case in West Africa occurred in June 1970. Since that time, many suspected cases have been reported and investigated clinically and by laboratory study. None have been found to be smallpox although, as described, eight have been proved to be caused by monkeypox virus. To determine whether any hidden focus of smallpox had persisted in West Africa, an intensive field survey was organized in 1975. Some five million children in 12 000 primary schools, maternity child health centres, and markets were seen by national and WHO survey teams looking for recent facial pockmarks. None of the children were found to have pockmarks resulting from a smallpox-like illness that had occurred after 1970, although many pockmarks were found among older children who had had smallpox prior to 1970. In addition, in the 8000 medical health units and markets in the countries, the teams specifically sought to learn of rumours of smallpox. All suspected smallpox cases reported to the teams were found to be chickenpox or other vesicular diseases.

In Zaire, Central Africa, where the last smallpox case was reported in 1970, 16 smallpox surveillance teams visit 4000 health units throughout the country once every six months to search for and investigate smallpox suspects. During the last five years, more than 500 specimens have been collected by these teams and tested by the WHO Collaborating Laboratories in Moscow, USSR and Atlanta, GA, USA. These surveillance activities in Zaire have resulted in the discovery of 12 cases of monkeypox in man but no smallpox cases.

These observations, together with the results of monkeypox surveys conducted five years after the outbreaks, provide increasingly convincing evidence that smallpox transmission in West and Central Africa has been interrupted and that the occurrence of monkeypox in man is extremely infrequent. The fact that man-to-man transmission of monkeypox virus appears to have taken place on two occasions is probably more of academic than of practical significance. Even if one assumes that both cases did occur as a result of man-to-man transmission, the rate of transmission among susceptible household contacts is so low that it would seem unlikely that persistent man-to-man transmission could be established. Notably, in the Aba area of Nigeria, where cases 7 and 8 occurred and where 80% of pre-school children had never been vaccinated (R. Netter, personal communication, 1976), monkeypox has not occurred in endemic form.

Although the animal reservoir of monkeypox virus is still unknown, it is assumed that such a reservoir does exist in West and Central Africa. For investigational purposes, Equateur Province, Zaire is of special interest since 8 of the 20 monkeypox cases were reported from this province. Whether the reservoir is monkeys or rodents, or perhaps other mammals, is uncertain. Laboratory studies show that the host range of monkeypox virus includes primates, rabbits, mice, and anteaters (16, 17).

Whitepox virus is indistinguishable from variola virus by currently available laboratory tests, but it is unknown whether it is capable of inducing infection in man and, if so, whether it can be transmitted from man to man. A generalized rash was produced in *C. aethiops* monkeys after subcutaneous or intraperitoneal introduction of high virus concentrations (10⁷ to 10⁸ PFU/ml) (J. Nakano, personal communication, 1976). These are unnatural routes of infection and no generalization can therefore be made about possible occurrences in nature. Furthermore, in

Equateur Province, where the presence of animal reservoirs of whitepox virus might be suspected, no smallpox-like disease except monkeypox has been

detected during the last 5-year period of surveillance. Therefore, whitepox virus and true variola virus may well behave differently in the human host.

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RÉSUMÉ

VIRUS « MONKEYPOX » ET « WHITEPOX » EN AFRIQUE OCCIDENTALE ET CENTRALE

Au cours de la période 1970–1975, 20 cas de maladie ressemblant à la variole ont été décelés dans des zones exemptes de variole de la forêt ombrophile tropicale de Sierra Leone, Libéria, Côte d'Ivoire, Nigéria, et Zaïre. Les recherches épidémiologiques et virologiques ont révélé que cette maladie était due à un poxvirus animal dit « monkeypox » qui appartient au groupe Orthopoxvirus. Dans deux cas seulement des adultes ont été frappés, tous les autres s'étant produits parmi des enfants non vaccinés dont quatre sont décédés. Les 20 cas étaient répartis dans 15 villages (à raison d'un cas unique dans 11 d'entre eux, de deux cas par village dans 3 autres et d'une poussée de trois cas dans le dernier).

En ce qui concerne les quatre villages où plusieurs cas se sont produits, dans deux la maladie s'est déclarée chez le second cas 9 à 10 jours après le premier, ce qui peut correspondre à une transmission d'homme à homme; dans les deux derniers, les malades ont présenté la maladie simultanément. Il y a eu 30 personnes non vaccinées en contact étroit avec les 20 cas; parmi elles, deux seulement ont contracté la maladie. Celle-ci se propage donc difficilement parmi les contacts sensibles. D'autres enquêtes spéciales effectuées dans quatre zones cinq ans après la survenue des atteintes humaines à monkeypoxvirus n'ont pas permis de déceler d'autres cas.

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